ESRF	**Experiment title:** FRANKFURT BAG: Quinol:fumarate reductases, membrane protein complexes from *Wolinella succinogenes* and *Campylobacter jejuni*	**Experiment number**: MX-135
Beamline: ID14-EH1	**Date of experiment**: from: 08-MAY-2003 8:00to: 09-MAY-2003 7:00	**Date of report**: 21-Sep-2003
Shifts: 3	**Local contact(s)**: Dr. Edward Mitchell	*Received at ESRF:*

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Report:

Quinol:fumarate reductase (QFR), couples the reduction of fumarate to succinate to the oxidation of quinol to quinone, in a reaction opposite to that catalysed by mitochondrial complex II (succinate dehydrogenase). QFR from the anaerobic bacterium *Wolinella succinogenes* consists of three protein subunits, FrdA, FrdB, and FrdC. Crystals of this bioenergetically important 130 kDa membrane protein complex diffract up to at least 1.8 Å and have previously been shown to have two different unit cells, both of the monoclinic space group $P2_1$. The unit cell of crystal form "A" is a = 85.2 Å, b = 189.0 Å, c = 117.9 Å, and β = 104.5º. Crystal form "B" has the unit cell dimensions a = 118.4 Å, b = 85.1 Å, c = 188.9 Å, β =96.5º. There are four complexes per unit cell and thus two complexes in the asymmetric units of both unit cells. Using data collected earlier at ESRF BM14 (cf. experimental reports for LS-1369), the structure of crystal form A has been solved by multiple isomorphous replacement and anomalous scattering (MIRAS) and refined to 2.2 Å resolution, and that of crystal form B has been solved by molecular replacement (MR) and refined to 2.33 Å resolution [1]. The structure of the enzyme in a third crystal form, form "C", with cell dimensions a = 81.1 Å, b = 290.2 Å, c = 153.6 Å, β =95.7º and four heterotrimeric QFR complexes in the asymmetric unit, has also been solved by molecular replacement [2], and refined at 3.1 Å resolution [3]. Mechanistically interesting variant enzymes have been

obtained by site-directed mutagenesis [2,3]. During the beam time available for this subproject, two data sets of form "A" crystals from a variant QFR enzyme could be collected (see Table for the statistics of the better data set) at T = 4°C from just one crystal each. The resulting structure is currently undergoing refinement.

Table. Diffraction data collected at ESRF ID14-EH1 on a crystal of a *W. succinogenes* QFR variant.

	resol. range [Å]	measured reflections	unique reflections	complete [%]	R_{sym} [%]
var_d03_4_3	50.0-2.50	566,353	123,934	100	9.4
	2.59-2.50		*12,380*	*100*	*41.8*

The remaining beam time was devoted to (ultimately unsuccesful) attempts to improve the previously recorded data set at 3.9 Å resolution of the QFR from *Campylobacter jejuni* (see Feb 2003 EH1 report).

References

[1] CRD. Lancaster, A Kröger, M Auer, H Michel (1999) *Nature* **402,** 377-385.
[2] CRD Lancaster, R Gross, A Haas, M Ritter, W Mäntele, J Simon, A Kröger (2000) *Proc. Natl. Acad. Sci. U.S.A.* **97**, 13051-13056.
[3] CRD Lancaster, R Gross, J Simon (2001) *Eur. J. Biochem.* **268**, 1820-1827.